**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

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| In re Application of: | ) | |
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| Jong Soo Woo et al. | ) | |
| | ) | Group Art Unit: 1655 |
| Serial No.: 10/576,196 | ) | |
| | ) | Examiner: Christopher R. Tate |
| Filed: October 21, 2004 | ) | |
| | ) | |
| For: ORAL MICROEMULSION COMPOSITION) | | |
| COMPRISING BIPHENYLDIMETHYL-) | | |
| DICARBOXYLATE AND SILYBIN ) | | |

**Commissioner for Patents**
**P. O. Box 1450**
**Alexandria, VA 22313-1450**

Sir:

DECLARATION UNDER 37 C.F.R. SECTION 1.132

I, Jong Soo WOO, being a citizen of the Republic of Korea and presently residing at Daewolmaeul 821-105, #914, Jeongja-dong, Jangan-gu, Suwon-si, Kyungki-do 440-300, Republic of Korea, do declare:

That I am one of the co-inventors of the invention disclosed in the above-identified application, and hence am fully familiar with the subject matter therein; and

That I have conducted a series of comparative experiments to demonstrate the unexpectedly improved therapeutic effects of the microemulsion compositions disclosed in the subject application, as follows.

**<Test 1> : Therapeutic effect on the liver damaged by carbon tetrachloride**

According to the procedure of Test Example 1 of the subject specification, the therapeutic effects on the liver damaged by $CCl_4$ of the inventive microemulsion formulations of **Examples 1 to 5** comprising as an active ingredient the combination of BDD and the *Carduus marianus* extract were examined together with five comparative cases of oral co-administration of two kinds of microemulsion formulations, one comprising BDD and another comprising the *Carduus marianus* extract (**Comparative Examples A to E**), based on the specifics shown in Table 1. The rats of **Negative control** were not treated with any therapeutic drug after the $CCl_4$ injection, and the rats of **Normal group** were not subjected to $CCl_4$ injection and no therapeutic drugs were administered thereto.

The serum ALT (alanine aminotransferase) and AST (aspartate aminotransferase) levels were measured, and the results are shown in Table 2.

**Table 1**

| | Drug composition | | Administration type |
|---|---|---|---|
| | BDD (mg) | *Carduus* extract (mg) | |
| Negative control | - | - | $CCl_4$ |
| Ex. 1 | 3 | 120 | (BDD+*Carduus*) composite administration |
| Ex. 2 | 3 | 60 | |
| Ex. 3 | 3 | 175 | |
| Ex. 4 | 25 | 175 | |
| Ex. 5 | 7 | 120 | |
| Comp. Ex. A | 3 | 120 | (BDD/*Carduus*) single formulation co-administration |
| Comp. Ex. B | 3 | 60 | |
| Comp. Ex. C | 3 | 175 | |
| Comp. Ex. D | 25 | 175 | |
| Comp. Ex. E | 7 | 120 | |
| Normal group | - | - | - |

**Table 2**

| Drug composition | | Examples | | Comparative Examples | |
|---|---|---|---|---|---|
| BDD (mg) | *Carduus* extract (mg) | ALT (SF U/ml) | AST (SF U/ml) | ALT (SF U/ml) | AST (SF U/ml) |
| 3 | 120 | 108.9±8.2 | 106.5±7.2 | 129.8±7.2 | 128.2±7.9 |
| 3 | 60 | 109.2±7.1 | 111.9±8.7 | 133.0±9.3 | 140.2±6.3 |
| 3 | 175 | 60.8±5.4 | 70.8±10.4 | 85.3±6.2 | 106.9±7.2 |
| 25 | 175 | 92.4±9.6 | 78.9±6.5 | 117.1±6.5 | 111.9±8.3 |
| 7 | 120 | 95.3±4.7 | 85.2±7.9 | 122.9±8.8 | 108.5±7.5 |

As shown in Table 2, the ALT and AST levels observed for the inventive microemulsion formulations were both lower by about 20 to 30 SF U/ml than those for comparative cases co-administered with the BDD and *Carduus marianus* extract microemulsions, which suggests that single administration of the inventive microemulsion formulation provides clearly superior therapeutic effects for liver diseases owing to synergistic effects of the two active ingredients, as compared to co-administration of the BDD and *Carduus marianus* extract microemulsions.

**<Test 2> : Verification of therapeutic synergistic effect using isobologram**

Using isobologram useful in the pharmaceutical field, the therapeutic synergistic effect of the inventive microemulsion formulation comprising as an active ingredient a combination of BDD and the *Carduus marianus* extract was verified. The procedure of <Test 1> was repeated employing the microemulsion formulations of Example 1 having the active ingredient's specifics shown in Table 3.

After the serum ALT and AST levels were measured, $RHT_{15}$ (reduction of hepatotoxicity), the amount (mg) of the active ingredient used to achieve 15% reduction in the ALT concentration, and $RHT_{25}$ (reduction of hepatotoxicity), the amount (mg) of the active ingredient used to achieve 25% reduction in the AST concentration were determined, and the results are shown in Table 4.

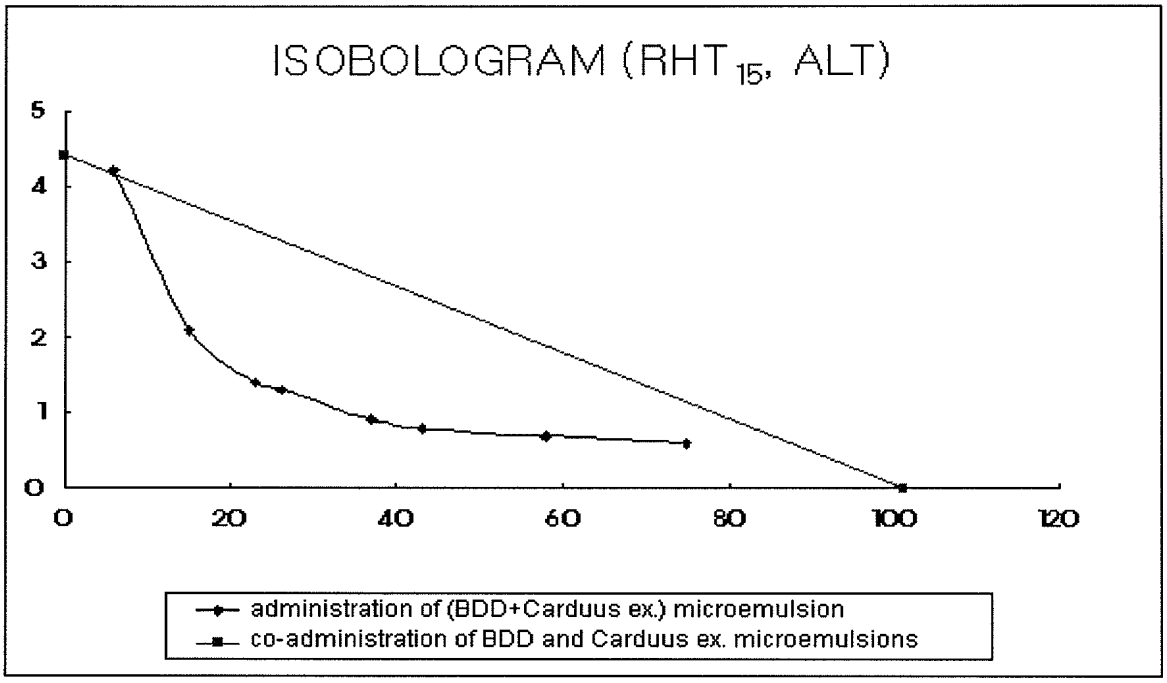
Further, isobolograms obtained from $RHT_{15}$ and $RHT_{25}$ values are depicted in the following figures.

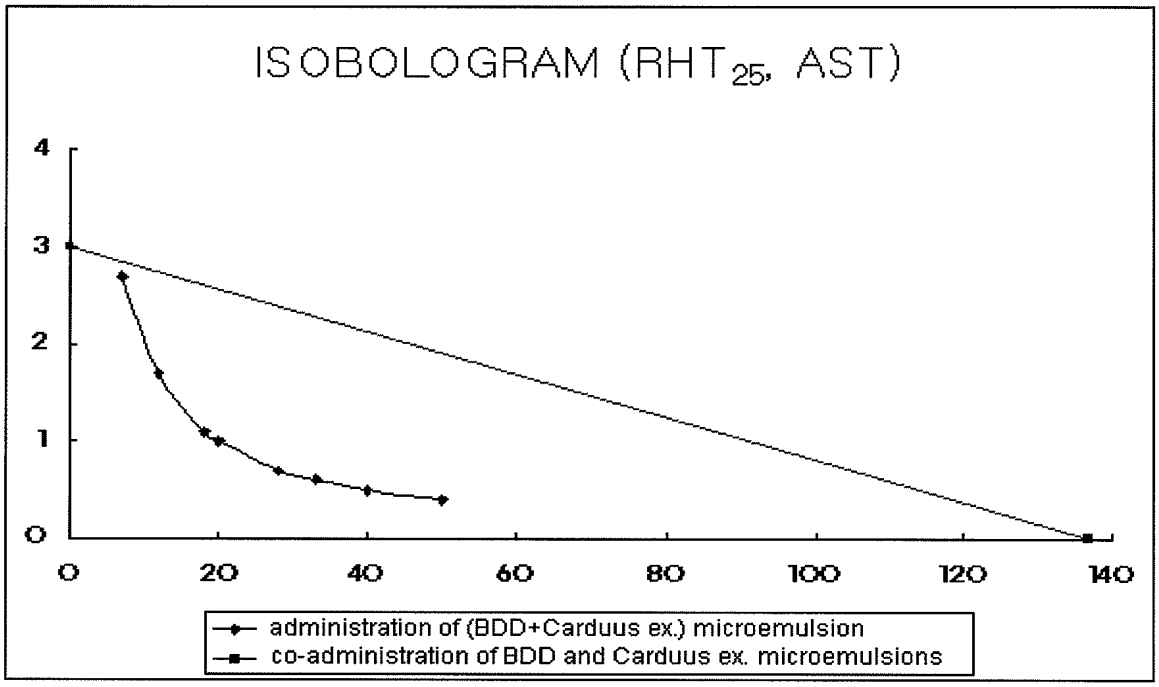
**Table 3**

| Group | | Drug composition | | Group | | Drug composition | |
|---|---|---|---|---|---|---|---|
| | | BDD (mg) | *Carduus* extract (mg) | | | BDD (mg) | *Carduus* extract (mg) |
| **2** | 2-1 | 1.5 | | **7** | 7-1 | 0.5 | 10 |
| | 2-2 | 3 | | | 7-2 | 1 | 20 |
| | 2-3 | 6 | | | 7-3 | 2 | 40 |
| | 2-4 | 12 | | | 7-4 | 4 | 80 |
| **3** | 3-1 | | 87.5 | **8** | 8-1 | 0.5 | 20 |
| | 3-2 | | 175 | | 8-2 | 1 | 40 |
| | 3-3 | | 350 | | 8-3 | 2 | 80 |
| | 3-4 | | 700 | | 8-4 | 4 | 160 |
| **4** | 4-1 | 0.5 | 1 | **9** | 9-1 | 0.5 | 29 |
| | 4-2 | 1 | 2 | | 9-2 | 1 | 58 |
| | 4-3 | 2 | 4 | | 9-3 | 2 | 116 |
| | 4-4 | 4 | 8 | | 9-4 | 4 | 232 |
| **5** | 5-1 | 0.5 | 3.5 | **10** | 10-1 | 0.5 | 42 |
| | 5-2 | 1 | 7 | | 10-2 | 1 | 83 |
| | 5-3 | 2 | 14 | | 10-3 | 2 | 166 |
| | 5-4 | 4 | 28 | | 10-4 | 4 | 332 |
| **6** | 6-1 | 0.5 | 8 | **11** | 11-1 | 0.5 | 63 |
| | 6-2 | 1 | 16 | | 11-2 | 1 | 125 |
| | 6-3 | 2 | 32 | | 11-3 | 2 | 250 |
| | 6-4 | 4 | 64 | | 11-4 | 4 | 500 |

**Table 4**

| | | $RHT_{15}$ | | $RHT_{25}$ | |
|---|---|---|---|---|---|
| | | BDD (Y-axis) | *Carduus* extract (X-axis) | BDD (Y-axis) | *Carduus* extract (X-axis) |
| BDD microemulsion (Group **2**) | | 4.4 (**A**) | 0 | 3 (**A'**) | 0 |
| *Carduus* ex. microemulsion (Group **3**) | | 0 | 101 (**B**) | 0 | 137 (**B'**) |
| BDD+ *Carduus* ex. microemulsion (BDD:*Carduus* ex. ratio) | 1:2 (Group **4**) | 4.2 | 6 | 2.7 | 7 |
| | 1:7 (Group **5**) | 2.1 | 15 | 1.7 | 12 |
| | 1:16 (Group **6**) | 1.4 | 23 | 1.1 | 18 |
| | 1:20 (Group **7**) | 1.3 | 26 | 1.0 | 20 |
| | 1:40 (Group **8**) | 0.9 | 37 | 0.7 | 28 |
| | 1:58 (Group **9**) | 0.8 | 43 | 0.6 | 33 |
| | 1:83 (Group **10**) | 0.7 | 58 | 0.5 | 40 |
| | 1:125 (Group **11**) | 0.6 | 75 | 0.4 | 50 |




In Table 4, **A** and **A'** values mean the amounts (mg) of BDD used to achieve 15% reduction in the ALT concentration and 25% reduction in the AST concentration, respectively; and **B** and **B'** values mean the amounts of the

*Carduus marianus* extract used to achieve 15% reduction in the ALT concentration and 25% reduction in the AST concentration, respectively. In this regard, X- and Y-values on the straight lines appearing in the isobolograms which are obtained by plotting **A** and **B** values, and **A'** and **B'** values, respectively, are regarded as respective amounts of BDD and the *Carduus marianus* extract for the same therapeutic effect when they are co-administered.

Accordingly, it is confirmed from the curves in the above isobolograms, the real values measured for the inventive microemulsion formulations, located below the straight lines that synergistic effects between the two active ingredients are indeed significant.

As can be seen from the above results, it can be concluded that the inventive microemulsion compositions provide greatly improved therapeutic effects for liver diseases due to the complementary work of the two active ingredients having different working mechanisms, as compared to the co-administration of the microemulsion compositions comprising BDD and the *Carduus marianus* extract, respectively.

The undersigned declarant further declares that all statement made therein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of United States Code and that such willful false statements may jeopardize the validity of the instant patent application or any patent issuing thereon.

Dated: June 26, 2008 By: J. S. Woo

Jong Soo WOO